REMARKS/ARGUMENTS

Claims 1-7 are active in this case. The claims are generally amended to correct obvious grammatical errors and to overall improve readability. The changes are supported throughout the specification, e.g., on page 3 and the Figures.

As amended to objections noted on page 2 of the Action are no longer applicable.

No new matter is believed to have been added by these amendments.

The present invention is related to the analysis of genes and, in particular, expression of genes in prokaryotic organisms. It is common knowledge in the relevant art that expression of most genes, whether in prokaryotic or eukaryotic organisms, through messenger RNAs (mRNA). In eukaryotic organisms the mRNA molecules include what is termed a "poly A" tail A, which is a sequence of adenine nucleotides that get added to the 3' end of the RNA. Therefore, it is common to use the poly A sequence as a target to selectively purify mRNA from total RNA extracts prepared from eukaryotic cells so as to enrich the population of mRNA molecules. By doing this, the specificity and sensitivity increases dramatically while substantially reducing background. As this knowledge is so well-known in this field, a publication has not been provided. Of course, should the Office require one, Applicants would be happy to provide one or more copies of such.

As discussed on page 1 of the present application, unlike eukaryotic mRNA molecules, prokaryotic mRNA molecules do not have poly A sequences and therefore the ability to effectively measure and/or analyze gene expression is hampered by the inability to simply and directly purify mRNA from the crude pool of total RNAs obtained from prokaryotic cells. The present invention solves this problem.

The invention solves this problem by first removing ribosomal RNA from total RNA, which significantly enriches the mRNA molecules in the RNA extract. To these mRNAs, a

polyA tail is added, which is then used to direct complementary DNA (cDNA) synthesis. Adapters are then provided to either end of the cDNA, which provides a point of hybridization to two primers for directing PCR-mediated amplification of the cDNA. This amplified pool of cDNA can be isolated and purified and then used in assessing the expression of prokaryotic genes, for example, measuring the level of expression of one or more genes under certain cultivation conditions. For further guidance on this, see pages 3-4 and Figure 1. In addition, for reference to Claim 4, see Figure 2.

It is respectfully submitted that the art cited by the Office in the rejections set forth in the Official Action do not describe and, certainly, do not suggest the invention as claimed.

Claims 1, 2-3, and 4-9 have been rejected under 35 USC 103(a) in three separate rejections. However, all three of the rejections rely primarily on the disclosure of the Weissman patent (US 5,712,126) in view of the Wendisch publication (*Anal Bioch* 90:205 (2001). The Wiesman patent teaches a method of forming cDNA from eukaryotic messenger RNAs and subsequent manipulations, including adapter ligation and PCR amplification. As correctly noted by the Office, however, Wiesman does not describe conducting such a method on prokaryotic mRNAs. Thus, the Office has cited Wendisch, who teaches adding poly A tails to prokaryotic mRNA and purifying the mRNA by selectively binding to the added poly A sequence on an oligo (dT) column. However, what is missing from this combination of art is a teaching to first remove ribosomal RNA from total RNA obtained from a prokaryotic cell to obtain a fraction of the total RNA enriched in mRNA (see Claim 1). In fact, the art would not have been modified to do this because Wendisch requires that poly A precedes any purification or enrichment step as the methodology used is directed to that modification.

Accordingly, withdrawal of the rejection of Claim 1 is requested.

The next rejection applies to Claims 2 and 3 and combined disclosures of Alland and Shah to the Weissman and Wendisch disclosures above. The Examiner relies on the Alland and Shah publications to allege that it would have been obvious to remove the rRNA from the mRNA. Applicants disagree. Notably, Alland teaches a way to avoid issues relating to high levels of rRNA by making a differential cosmid library (term "differential expression using customized amplification libraries" DECAL) (col. 3, lines 16-26 and col. 7, lines 1-6). In other words, there is no selective removal of the rRNA from mRNA in a total RNA extract prior to forming a cDNA or conducting PCR as in the claims. More significant is the fact that Alland specifically teaches that his method is applicable to situations where polyadenylation of prokaryotic messenger RNA is not desired.

Therefore, as the method of Alland teaches a different approach to the ones of Weismann and Wendisch they would not have been combined because Alland teaches a method for polyA (-) situations where the Weisman and Wendisch references would be applicable to polyA(+) situations (as is clear from the obviousness rejection raised on Claim 1). Moreover, neither method would have been modified to incorporate each others teachings because Alland specifically teaches away from the method alleged to be apparent from the combination of Weismann and Wendisch as well as the invention claimed, which requires the addition of poly A sequences to prokaryotic mRNA.

Even more significant is the fact, that these references do not describe the claimed method in which the rRNA is removed before manipulation of the mRNA and is the exact opposite of the teachings of Alland, who requires co-manipulation of the rRNA and mRNA (they make cosmids and then keep only those RNA fractions they want).

The addition of Shah to these references doesn't alleviate the problems with making a case for obvious as discussed above. Notably, while Shah does discuss the usefulness of magnetic beads as a tool for isolation, what Shah when combined with Alland, Weismann

and Wendlich fail to teach is to remove rRNA from mRNA and then subsequently attach polyA to mRNA for further manipulation and characterization.

Accordingly, Claims 2 and 3 would not have been obvious in view of the cited art and as such withdrawal of this rejection is requested.

As to the rejection of Claims 4-9 in view of Weissman, Wendisch and Belyavsky, this rejection is also not sustainable. As discussed above in the context of the rejection of Claim 1, the art fails to teach removing ribosomal RNA from total RNA obtained from a prokaryotic cell to obtain a fraction of the total RNA enriched in mRNA (see Claim 1). In fact, the art would not have been modified to do this because Wendisch requires that poly A precedes any purification or enrichment step as the methodology used is directed to that modification. The Belyavsky publication is silent with respect to these claimed features and provides no reasons to modify the Weissman and Wendisch references in a manner that would lead one to the claimed invention.

Withdrawal of the rejection of Claims 4-9 is also requested.

The rejection of Claims 1-9 under 35 USC § 112, second paragraph is no longer applicable in light of the amendments submitted herein. Specifically,

Claim 1 has been provided with specific steps in conducing the process (i.e., "adding," synthesizing," "attaching," etc.);

Claim 1 has also been amended to correlate the recovery of the amplified cDNA with analyzing gene expression as set forth in the preamble,

the aspect of hybrid formation in Claims 2 and 3 is believed to be clear, e.g., the first and third polynucleotides form a hybrid with the 16S rRNA and the second and third polynucleotides form a hybrid with the 23S rRNA; and

where the tag substance is attached has been clarified.

Withdrawal of this rejection is requested.

A Notice of Allowance for all pending claims is earnestly solicited.

Should the Examiner deem that any further action is necessary to place this application in even better form for allowance, she is encouraged to contact Applicants' undersigned representative.

Respectfully submitted,

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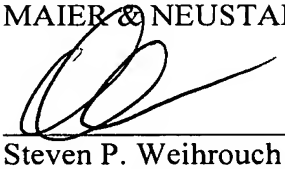

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